

WHAT IS CLAIMED IS:

1. A method of detecting or identifying an analyte comprising:
 - a) exposing less than about 10^3 molecules of an analyte to at least one Raman-active surface;
 - 5 b) irradiating the interface between at least one molecule and said surface with a laser beam at a first wavelength, such that said molecule produces a spontaneous Stokes Raman emission at a second wavelength and a spontaneous anti-Stokes Raman emission at a third wavelength;
 - c) substantially simultaneously with b), irradiating the interface
10 between said molecule and the surface with a second beam at said second wavelength, such that the intensity of said anti-Stokes Raman emission from said molecule at said third wavelength increases; and
 - d) detecting or identifying said analyte by detecting or identifying the intensity change of said anti-Stokes emission from the interface at said third
15 wavelength following b) and c) with a Raman detection unit.
2. The method of claim 1, comprising exposing less than about 10^2 molecules of an analyte to at least one Raman-active surface.
3. The method of claim 1, comprising exposing less than about 10 molecules of an analyte to at least one Raman-active surface.
- 20 4. The method of claim 1, comprising exposing a single molecule of an analyte to at least one Raman-active surface.
5. The method of claim 1 further comprising moving said analyte through a channel.
6. The method of claim 5 further comprising detecting and identifying
25 said analyte in aqueous media.
7. The method of claim 1 wherein said surface is provided by a member selected from the group consisting of a silicon substrate coated with a metal or conductive material; a metallic or conductive nanoparticle; an aggregate of metallic or conductive nanoparticles; a colloid of metallic or conductive nanoparticles; and
30 combinations thereof.

8. The method of claim 1, wherein the analyte is selected from the group consisting of an amino acid, peptide, polypeptide, protein, glycoprotein, lipoprotein, nucleoside, nucleotide, oligonucleotide, nucleic acid, sugar, carbohydrate, oligosaccharide, polysaccharide, fatty acid, lipid, hormone, metabolite, cytokine, chemokine, receptor, neurotransmitter, antigen, allergen, antibody, substrate, metabolite, cofactor, inhibitor, drug, pharmaceutical, nutrient, prion, toxin, poison, explosive, pesticide, chemical warfare agent, biohazardous agent, bacteria, virus, radioisotope, vitamin, heterocyclic aromatic compound, carcinogen, mutagen, narcotic, amphetamine, barbiturate, hallucinogen, waste product and contaminant.
9. The method of claim 8, wherein the analyte is a nucleoside, nucleotide, oligonucleotide, nucleic acid, amino acid, peptide, polypeptide or protein.
10. The method of claim 9, wherein said analyte is a nucleic acid.
11. The method of claim 1, wherein said analyte is closely associated with a Raman-active surface.
12. The method of claim 1, wherein the Raman-active surfaces are covalently modified with organic compounds.
13. The method of claim 5, wherein said channel is selected from the group consisting of a microfluidic channel, a nanochannel, a microchannel, and combinations thereof.
14. The method of claim 13, wherein said nanoparticles are between about 1 nm and 2 μm in size.
15. The method of claim 13, wherein the size of said nanoparticles is selected from the group consisting of about 10 to 50 nm, about 50 to 100 nm, about 10 to 100 nm, about 100 nm and about 200 nm.
16. The method of claim 7, wherein the metal is selected from the group consisting of gold, silver, copper, platinum, aluminum, and combinations thereof.
17. The method of with claim 1, wherein said Raman detection device is selected from the group consisting of photodiodes, avalanche-photodiodes, charge coupled devices (CCDs), charge coupled device arrays, complementary metal oxide semiconductor (CMOS) arrays, and combinations thereof.

18. The method as claimed in claim 1 further comprising comparing the detected intensity for the analyte to a previously identified analyte so that the identity of the analyte can be determined.

19. The method of claim 1 wherein the method has an optical cross
5 section of at least about 10^{-22} cm² per molecule.

20. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-20} cm² per molecule.

21. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-19} cm² per molecule.

10 22. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-18} cm² per molecule.

23. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-17} cm² per molecule.

15 24. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-16} cm² per molecule.

25. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-15} cm² per molecule.

26. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-14} cm² per molecule.

20 27. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-13} cm² per molecule.

28. The method of claim 1 wherein the method has an optical cross section of at least about 10^{-12} cm² per molecule.

25 29. The method of claim 1, wherein said analyte is labeled with one or more distinguishable Raman labels.

30. The method of claim 1, further comprising imposing an electric field to move said analyte through said channel.

31. The method of claim 1, wherein each type of analyte produces a unique Raman signal.

32. A device for detecting less than about 10^3 molecules of an analyte, said device comprising:

(a) means for producing a first beam of electromagnetic radiation at a first wavelength;

5 (b) means for producing a second beam of electromagnetic radiation at a second wavelength, said second wavelength differing from said first wavelength;

(c) a sample cell;

(d) means for introducing said analyte and a Raman-active surface to said sample cell;

10 (e) optics for focusing said first beam and said second beam onto an interface between said analyte and said Raman active surface; and

(f) means for detecting the intensity of light emitted from the interface between the analyte and the Raman-active surface, positioned to receive said emission.

15 33. The device in accordance with claim 32, wherein said means for producing a first beam of electromagnetic radiation and said means for producing a second beam of electromagnetic radiation comprise at least one pulsed laser.

34. The device in accordance with claim 32, wherein said optics comprises a microscope objective lens, a mirror, a prism, or combinations thereof.

20 35. The device in accordance with claim 32, wherein said sample cell comprises:

(a) a sample cell body of a material that isolates said sample from ambient air;

25 (b) a window in said sample cell body of a material that is transparent to electromagnetic radiation; and,

(c) at least one port for introducing and removing said analyte and optionally the Raman-active surface.

36. The device in accordance with claim 32, wherein said means for introducing an analyte and a Raman-active surface to said sample cell comprises a

channel, a microfluidic device, an integrated chip, or a microelectricalmechanical system (MEMS).

37. The device in accordance with claim 32, wherein said means for detecting the intensity of light emitted from the interface is a differential photo-
5 detecting device selected from the group consisting of photodiodes, avalanche-
photodiodes, charge coupled devices (CCDs), charge coupled device arrays, complementary metal oxide semiconductor (CMOS) arrays, and combinations thereof.

38. A device for detecting less than about 10^3 molecules of an analyte,
10 said device comprising:

- a) a sample cell in fluid communication with said first and second channels;
- b) at least one Raman-active surface in said sample cell;
- c) a laser; and
- 15 d) a surface enhanced, coherent anti-Stokes Raman detector operably coupled to said sample cell.

39. The device of claim 38, wherein said Raman detector is selected from the group consisting of photodiodes, avalanche-photodiodes, charge coupled devices (CCDs), charge coupled device arrays, complementary metal oxide
20 semiconductor (CMOS) arrays, and combinations thereof.

40. The device of claim 38, wherein said Raman detector is operably coupled to a data processing unit.

41. The device of claim 38, wherein said data processing unit comprises a computer.

25 42. The device of claim 38, further comprising a channel.

43. The device of claim 38, further comprising a first electrode and a second electrode, said electrodes to move analytes through said channel.

44. The device of claim 38, wherein said channel is a nanochannel, a microchannel, or a microfluidic channel.

45. The device of claim 38, wherein the Raman-active surface is provided by a member selected from the group consisting of a silicon substrate coated with a metal or conductive material; a metallic or conductive nanoparticle; an aggregate of metallic or conductive nanoparticles; a colloid of metallic or conductive nanoparticles; and combinations thereof.

46. The device of claim 45, wherein the metal comprises silver, gold, platinum, copper and/or aluminum.

47. The device of claim 38, further comprising a flow through cell operably coupled to the Raman detector, wherein flow passes by the Raman-active surface inside the sample cell.

48. The device of claim 38, wherein the Raman-active surface is incorporated into a channel, sample cell, microfluidic device, an integrated chip, or micro-electro-mechanical system (MEMS).

49. The device of claim 38, wherein said Raman detector is selected from the group consisting of photodiodes, avalanche-photodiodes, charge coupled devices (CCDs), charge coupled device arrays, complementary metal oxide semiconductor (CMOS) arrays, and combinations thereof.